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(54) Title: A PROCESS FOR SEQUENCE SATURATION MUTAGENESIS (SESAM)

(57) Abstract: A process for the mutagenesis of a double-stranded polynucleotide sequence (master sequence) of n base-pairs having a (+)-strand and a complementary (-)-strand comprising the steps (i) creation of a collection of single-stranded fragments of the (+)-strand of the master sequence wherein all members of the collection have the same 5'-terminus and have a deletion in the 3'-terminus such that the collection represents (+)-strands with a length of n-1, n-2, n-3, nucleotides; (ii) introduction of at least one universal or degenerate nucleotide at the 3'-terminus of the (+) strand produced in step (i); (iii) elongation of the (+)-strand produced in step (ii) to the full length of the master sequence using the (-)-strand or fragments thereof as a template strand for the elongation; (iv) synthesis of a (-)-strand by using the (+)-strand produced in step (iii) as a template strand thereby effecting mutations in the (-)-strand at the positions of the previous universal or degenerate nucleotides compared to the master sequence.

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